

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Lgr4/GPR48 in Lgr4-transfected cell-based ELISA. Stains human Lgr4/GPR48 transfected but not irrelevant transfected cells in flow cytometry.
Source	Monoclonal Mouse IgG _{2B} Clone # 852229
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	NS0 mouse myeloma cell line transfected with human Lgr4/GPR48 Met1-Asp951 Accession # Q9BXB1
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

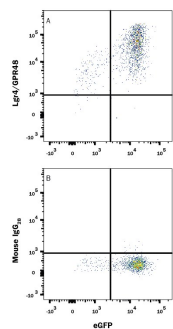
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Flow Cytometry	0.25 µg/10 ⁶ cells	See Below
Immunocytochemistry	8-25 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	
Blockade of Receptor-ligand Interaction	In a functional flow cytometry test, 2.5 µg/mL of Mouse Anti-Human Lgr4/GPR48 Monoclonal Antibody completely blocks binding of biotinylated Recombinant Human R-Spondin 4 (100 ng/mL) to HEK293 human embryonic kidney cell line transfected with human Lgr4/GPR48.	

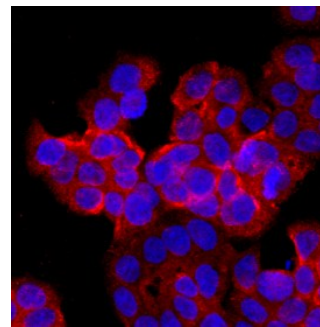
DATA

Flow Cytometry



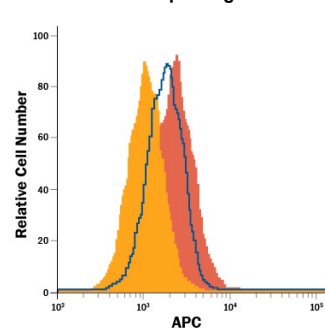
Detection of Lgr4/GPR48 in HEK293 Human Cell Line Transfected with Human Lgr4/GPR48 and eGFP by Flow Cytometry. HEK293 human embryonic kidney cell line transfected with human Lgr4/GPR48 and eGFP was stained with either (A) Mouse Anti-Human Lgr4/GPR48 Monoclonal Antibody (Catalog # MAB7750) or (B) Mouse IgG_{2B} Flow Cytometry Isotype Control (Catalog # MAB0041) followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B). View our protocol for [Staining Membrane-associated Proteins](#).

Immunocytochemistry



Lgr4/GPR48 in HT-29 Human Cell Line. Lgr4/GPR48 was detected in immersion fixed HT-29 human colon adenocarcinoma cell line using Mouse Anti-Human Lgr4/GPR48 Monoclonal Antibody (Catalog # MAB7750) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cell surfaces. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

Blockade of Receptor-ligand Interaction



R-Spondin 4 Binding to Lgr4/GPR48-transfected HEK293 Human Cell Line is Blocked by Human Lgr4/GPR48 Antibody. In a functional flow cytometry test, biotinylated Recombinant Human R-Spondin 4 (100 ng/mL) binds to HEK293 human embryonic kidney cell line transfected with human Lgr4/GPR48 (dark orange histogram). Binding is completely blocked (light orange histogram) by 2.5 µg/mL of Mouse Anti-Human Lgr4/GPR48 Monoclonal Antibody (Catalog # MAB7750). Mouse IgG_{2B} Isotype Control (Catalog # MAB0041) at 2.5 µg/mL was used as a control (blue line). Cells were stained with Streptavidin-APC (Catalog # F0050).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Lgr4 (Leucine-rich repeat GPR 4), also known as GPR48 (G-Protein-coupled Receptor 48), is a seven-transmembrane glycoprotein receptor in the Lgr family of cell surface receptors (1, 2). While this family includes receptors for hormones such as LH, FSH, TSH, and HCG, the subfamily comprising Lgr4, Lgr5, and Lgr6 are G-protein-independent mediators of the potentiating effect of R-Spondins on Wnt signaling (1-6). Lgr4 binds and forms complexes with R-Spondins, Frizzled Wnt receptors and LRP Wnt co-receptors (5). It acts at least in part by enhancing Wnt-dependent LRP phosphorylation, internalization of LRPs, and accumulation of β -catenin (3, 4). Human Lgr4 cDNA encodes 951 amino acids (aa), including a long N-terminal Extracellular Domain (ECD, aa 25-544) with 16-17 LRR domains that mediate ligand interaction (1). The LRR-containing ECD of human Lgr4 shares 93% aa sequence identity with mouse, rat and bovine Lgr4, and 50-60% aa identity with human Lgr5 and Lgr6. Lgr4 is widely expressed in both embryo and adult. Expression of Lgr4 mRNA in adult humans is highest in pancreas, followed by liver, heart, muscle, brain, and placenta (1). In rodents, embryonic and adult expression includes liver, kidney, adrenals, bone/cartilage, and heart (2, 7-9). Lgr4 deletion in the mouse affects development in areas of expression, for example, inhibiting fetal liver definitive erythropoiesis (9). Deletion of Lgr4 specifically from stem and progenitor cells in intestinal crypts induces loss of crypts due to insufficient Wnt signaling (5, 6). Lgr4 may be over-expressed in carcinomas and may promote invasiveness and metastasis by down-regulating p27^{Kip1} expression (10).

References:

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